Adaptive changes in the lipids of higher-plant membranes

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The membrane lipids of higher plants are altered by the same diverse environmental factors as those that effect other poikilotherms (i.e., temperature, water-stress, nutritional deprivation and salt-stress). In addition, pollutants (including herbicides) have been noted, in certain cases, to cause significant alterations of plant membrane lipids. However, perhaps the most noticeable and spectacular effects concern the degree of unsaturation in membranes from these tissues is relatively less fluid than those from leaves. However, even in leaves, growth at low temperatures was found to increase the degree of unsaturation of the phospholipid fatty acids (e.g. Chapman & Barber, 1980). This phenomenon has frequently been connected with the property of membrane fluidity (Raison, 1980). The effects observed are usually largest in non-photosynthetic tissues because the degree of unsaturation in membranes from these tissues is relatively less than those from leaves. However, even in leaves, growth at low temperatures was found to increase the degree of unsaturation of the phospholipid fatty acids (e.g. Chapman & Barber, 1980).

Temperature effects

Many workers have shown that temperature has an effect on the relative proportions of saturated and unsaturated fatty acids in the membrane lipids of plant tissues and in isolated plant cells (cf. Hitchcock & Nichols, 1971; Harwood, 1975; Mazliak et al., 1980; Wintermans & Kuiper, 1982). This phenomenon has frequently been connected with the property of membrane fluidity (Raison, 1980). The effects observed are usually largest in non-photosynthetic tissues because the degree of unsaturation in membranes from these tissues is relatively less than those from leaves. However, even in leaves, growth at low temperatures was found to increase the degree of unsaturation of the phospholipid fatty acids (e.g. Chapman & Barber, 1980).

There has been some speculation as to the reason for increased desaturation in plants at low temperatures. Theories can be grouped under two headings: those that suggest changes in the activity or amounts of the desaturase enzymes themselves or, alternatively, a proposal by Harris & James (1969) that the oxygen concentration of the medium plays a key role in regulating unsaturation. Support for the latter idea has come from the recent experiments by Rebelle et al. (1980) with plant cells in culture. They found that sycamore cells would produce exactly the same fatty acid proportions at 15°C and 25°C provided that the oxygen concentration was held constant. Furthermore, the linoleate concentration could be increased from 15% to 45% by increasing the oxygen content in the medium of cells grown at 25°C.

Continued growth at a constant temperature produces one type of lipid pattern, but plants are also exposed to temperatures outside the normal range they have been accustomed to. Considerable attention has been devoted to the phenomenon of 'frost hardness' (with all of its important agricultural
implications). Frost hardening in plants is often accompanied by an increased level of phosphatidycholine and phosphatidyl-
ethanolamine (e.g. Smolenska & Kuiper, 1977; Clarkson et al., 1980). In many cases, these changes in the relative quantities of acyl lipids are much more obvious than alterations in degrees of unsaturation. Indeed, in many instances it has proved difficult to correlate the degree of unsaturation with the frost resistance of certain varieties of a given plant (e.g. de la Roche et al., 1975; Raison, 1980). Vigh (1982) has also shown some large changes in the lipid composition but not in the unsaturation of thylakoid membranes isolated from the leaves of frost-resistant and frost-sensitive wheat exposed to hardening conditions. These effects are also carried over into the chilling resistance of certain seeds such as those of *Pinus*. Thus, Palacios-Alaix *et al.* (1982) found that the phospholipid content and unsaturation index were increased in seeds germinated at low temperatures.

Recently, Kinney *et al.* (1982) have attempted to discover why frost-stressed plants accumulate increased levels of phospholipids. Their experiments with rice roots have shown an increase in the incorporation of choline and ethanolamine into their respective lipids by the CDP-base pathway. The activity of the terminal enzymes of this pathway (i.e. cholinephospho-
transferase and ethanolaminephosphotransferase) were 5-fold higher in roots from rice plants grown at 5°C in comparison with those grown at 20°C.

Salt stress and other effects of minerals

Lipids that are important in the regulation of membrane permeability may play a role in the phenomenon of salt tolerance in plants. A number of experiments have demonstrated a salt-induced decrease of chloroplast lipids, sometimes accompanied by a decrease in the degree of fatty acid unsaturation (Müller & Santarius, 1978; Harzallah-Skhirí *et al.*, 1980). Other studies have demonstrated a decrease in all the acyl lipids of salt-stressed plants (Erdeí *et al.*, 1980; Zarrouk & Cherif, 1981). However, it seems unlikely that salt-tolerance in *Plantago* spp. is due to the changes in phospholipid concen-
tration (Tuiver *et al.*, 1982). In sunflower leaves and roots, NaCl treatment decreased the total lipid content of leaves, CaSO₄ treatment caused an increase (Bettaieb *et al.*, 1980). Plants that differ in their requirements for calcium have been categorized as calcifuge [plants thriving in soils poor in Ca(NO₃)₂, e.g. *Lupinus luteus*] and calcilcole [plants thriving in soils rich in Ca(NO₃)₂, e.g. *Vicia faba*]. Calcifuge plant roots have a higher content of acidic lipids and saturated fatty acids than calcilcole plants (Rossignol, 1976). In calcilcole plants calcium was found to inhibit phospholipid synthesis and both cholinephosphotransferase and ethanolaminephosphotransferase were sensitive to this inhibition (Orursel, 1979). Calcifuge plants, on the other hand, show increased rates of fatty acid desaturation when grown in calcium-enriched media (Citharel *et al.*, 1982).

Another environmental factor that can be mentioned at this stage is water stress. Adaptation of plants to drought has been shown to lead to an increase in phospholipid content and phospholipid synthesis. Changes in the fatty acid constituents of the phospholipids and sulpholipids were also noted (cf. Tuiver *et al.*, 1982). Water stress has also been found to change the ratio of sterols to acyl lipids in oat root membranes (Liljenberg & Kates, 1982) and to lower the levels of α-linolenate and *trans*-hexadec-3-enoate in cotton leaves (Pham Thi *et al.*, 1982).

**Influence of light on plant lipids**

Since chloroplastic membranes contain high concentrations of lipids then their development has a considerable effect on the composition of photosynthetic tissues. The effect of light on leaf lipids has been studied most often by growing plants in the dark and then ‘greening’ the etiolated tissues under defined conditions. Etioplasts contain lower concentrations of α-linolenate than chloroplasts and have a strikingly different morphological appearance, with a very prominent prolamellar body. Although the relative proportions of acyl lipids are rather similar in the two organelles, the phosphatidylglycerol of etioplasts is virtually devoid of *trans*-hexadec-3-enoate (Harwood, 1980). Dark-grown lipids do, however, contain reduced amounts of lipids in terms of tissue fresh weight.

Usually, monocotyledons have been used for studies because the leaves of dicotyledons do not expand well in the dark. Greening involves a net synthesis of galactolipids in *Kalanchoe* *aerophila* (Thomas & Stewart, 1970) and, in barley, the production of diacylgalactosylglycerol containing linolenic acid (Appelqvist *et al.*, 1968). Some workers believe there may be a special relationship between sulpholipid and chlorophyll during greening (cf. Harwood, 1980). In a study of the effect of blue light on the conversion of barley etioplasts into chloroplasts, Tevini (1977) noted an initial drop in the percentage of

**Table 1. Changes in the lipid composition of etiolated barley leaves as they are exposed to white light**

Barley plants were grown at 20°C in darkness for 7 days. They were then exposed to white light (200μE·s⁻¹·m⁻²) for the time indicated. Lipids were extracted and analysed as detailed in Wharfe & Harwood (1978). Abbreviations used: C₁₆:₀ palmitic acid; C₁₆:₁,₁ hexadecanoic acids; C₁₈:₀ stearic acid; C₁₈:₁ oleic acid; C₁₈:₂ linoleic acid; C₁₈:₃ α-linolenic acid; n.d., not detected: tr, trace amount; *, Palmitoleic acid; †, 1% palmitoleic acid, 2% *trans*-hexadec-3-enoic acid; ‡, 1% palmitoleic acid, 27% *trans*-hexadec-3-enoic acid.

<table>
<thead>
<tr>
<th>Lipid</th>
<th>Amount (μg/mg fresh wt)</th>
<th>Acyl lipids (%)</th>
<th>Fatty acid composition (% of total)</th>
</tr>
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<tbody>
<tr>
<td></td>
<td></td>
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<td>C₁₆:₀</td>
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<tr>
<td>Etiolated tissue</td>
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<tr>
<td>Phosphatidylcholine</td>
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<tr>
<td>Phosphatidylcholine</td>
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<td>26</td>
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<tr>
<td>Diacylgalactosylglycerol</td>
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<tr>
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1983
diacylgalactosylglycerols. This was followed in the 8–16 h period by an increase in typical chloroplast lipids, such as diacylgalactosylglycerol, diacylgalactosylglycerol and phosphatidylglycerol. Similar data have been obtained by other workers (cf. Harwood, 1980; Table 1).

In an effort to differentiate light effects per se from those connected with light-induced chloroplast formation, Gemmrich (1982) has studied the lipids of green light-grown cultures, non-green light-grown cultures and dark-grown cultures of *Ricinus communis*. His results showed clearly that the light-induced changes were associated with thylakoid membranes.

The connection of trans-hexadec-3-enolate with active thylakoid membranes has led to speculation as to its role in photosynthesis. In particular, it was proposed that the acid was made specifically for granal stacking (Tuquet et al., 1977) and its synthesis was correlated with the exposure of etiolated leaves to monochromatic wavelengths that also induced stacking (Trémolieres et al., 1979). However, experiments with different treatments (Percival et al., 1979) and chlorophyll-less mutants (Bolton et al., 1978; Selstam, 1980) have largely discounted this theory (cf. Trémolieres et al., 1982). There now seems a strong possibility that phosphatidylglycerol containing trans-hexadec-3-enolate may be associated with the oligomeric organization of the light-harvesting chlorophyll–protein complex.

Although light effects seem to be associated only with the development and function of photosynthetic membrane lipids, light has been shown to stimulate the activity of certain chloroplastic enzymes. For example, acetyl-CoA carboxylase, fatty acid synthesis *de novo* and phosphatidylglycerol synthesis *de novo* are all greatly stimulated in chloroplast fractions. Furthermore, the use of light qualities (such as far-red wavelengths) that do not allow normal chloroplast differentiation also leads to an unusual pattern of lipid accumulation and synthesis (Table 2).

Although the effects of light that have been discussed above apply to acyl lipids, it should not be forgotten that light also affects the synthesis of other plant lipids, notably pigments. Wax components (Avato et al., 1980) and prenylquinones (Lichtenthaler, 1980) are included amongst those lipids whose amounts are altered by light exposure or intensity.

**Other environmental factors that alter plant membrane lipids**

Although not always coming under the heading of adaptation, plant membrane lipids may be altered by a number of other environmental factors. These include nutritional status, soil acidity and fungal infections. It should also be borne in mind that several herbicides act by inhibiting lipid synthesis. For further details the reader is referred to Hitchcock & Nichols (1971), Harwood (1980), Mazliak et al. (1980) and Wintermans & Kuiper (1982).

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**Table 2. Effect of far-red light on the incorporation of radioactivity from [1-14C]acetate into lipids of greening barley leaves**

Plants were grown in the dark for 7 days. Leaves were detached and exposed either to white light (200 µE·m⁻²·s⁻¹) or to >700 nm light (46 µE·m⁻²·s⁻¹) for 8 h at 20°C in the presence of 1 µCi of [1-14C]acetate/leaf. See Wharfe & Harwood (1978) for further details of the methods used. Abbreviations used: DGG, diacylgalactosylglycerol; DDG, diacyldigalactosylglycerol; SQDG, diacylsulphoquinovosylglycerol; PC, phosphatidylcholine; PE, phosphatidylethanolamine; PG, phosphatidylglycerol; tr, trace amount. For fatty acid abbreviations see the legend to Table 1.

| Acyl lipid labelling (% of total [%14C]acyl groups) | DGG | DDG | SQDG | PC | PE
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<td>7</td>
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<td>9</td>
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<table>
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<th>C₁₆:₀</th>
<th>C₁₈:₀</th>
<th>C₁₆:₁</th>
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</thead>
<tbody>
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<td>White light illumination</td>
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<td>3</td>
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